

**Amendments to the Claims:**

This listing of claims will replace all prior versions, and listings, of claims in the application:

**Listing of Claims:**

1-32. (Canceled)

33. (Withdrawn-currently amended) A method for isolating a microRNA of interest from a sample comprising the microRNA of interest; the method comprising:

a) providing a sample comprising the microRNA sequence of interest;

b) providing a capture probe comprising:

i) a first adapter segment having a first adapter segment sequence, the first adapter segment comprising a 3' end and a 5' end;

ii) a second adapter segment having a second adapter segment sequence, the second adapter segment comprising a 3' end and a 5' end; and

iii) a microRNA binding segment having a microRNA binding segment sequence;

where the microRNA binding segment is substantially complementary to, and capable of hybridizing to, one or more than one microRNA of interest by Watson-Crick base pairing;

where the 5' end of the first adapter segment is connected to the 3' end of the microRNA binding segment; and

where the 3' end of the second adapter segment is connected to the 5' end of the microRNA binding segment;

c) providing a first linker, wherein the first linker has a first linker sequence, comprises a 3' end and a 5' end, and is substantially complementary to, and capable of hybridizing to, the first adapter segment of the capture probe ~~and a second linker;~~



d) providing a second linker, wherein the second linker has a second linker sequence, comprises a 3' end and a 5' end, and is substantially complementary to, and capable of hybridizing to, the second adapter segment of the capture probe;

[d]e) combining the sample, the capture probe, the first linker and the second linker in a solution;

[e]f) allowing the first linker to hybridize with the first adapter segment, the microRNA of interest to hybridize with the microRNA binding segment, and the second linker to hybridize with the second adapter segment;

[f]g) ligating the 3' end of the first linker ~~that is hybridized to the first adapter segment~~ to the 5' end of the microRNA of interest ~~that is hybridized to the microRNA binding segment~~, and ligating the 3' end of the microRNA of interest ~~that is hybridized to the microRNA binding segment~~ to the 5' end of the second linker ~~that is hybridized to the second adapter segment~~, thereby producing a complex defined as a strand of first linker, microRNA of interest and second linker that have been ligated together (ligated first linker- microRNA of interest-second linker) and that is hybridized to the capture probe;

[g]h) dehybridizing the capture probe from the strand of the ligated first linker-microRNA of interest-second linker; and

[h]i) purifying the ligated first linker- microRNA of interest-second linker, thereby purifying the microRNA of interest ~~that has been dehybridized from the capture probe;~~

~~where the microRNA of interest has a microRNA of interest sequence, and comprises a 3' end and a 5' end;~~

~~where the microRNA of interest is substantially complementary to, and capable of hybridizing to, the microRNA binding segment of the capture probe by Watson-Crick base pairing;~~

~~where the first linker has a first linker sequence, and comprises a 3' end and a 5' end;~~



~~where the first linker is substantially complementary to, and capable of hybridizing to, the first adapter segment of the capture probe by Watson-Crick base pairing;~~

~~where the second linker has a second linker sequence, and comprises a 3'-end and a 5'-end; and~~

~~where the second linker is substantially complementary to, and capable of hybridizing to, the second adapter segment of the capture probe by Watson-Crick base pairing;~~

~~where purifying the ligated first linker-microRNA of interest-second linker comprises:~~

~~——— i) applying DNAase to a solution containing the ligated first linker-microRNA of interest-second linker to destroy any DNA present in the solution, where the first linker, or the second linker, or both the first linker and the second linker comprise nuclease-resistant nucleotides, or comprise nucleotides with a phosphothioate backbone that render the first linker, or the second linker, or both the first linker and the second linker resistant to nuclease degradation; or~~

~~——— ii) circularizing the ligated first linker-microRNA of interest-second linker and treating the solution containing the circularized ligated first linker-microRNA of interest-second linker with one or more than one exonuclease.~~

34. (Withdrawn) The method of claim 33, where the sample further comprises one or more than one substance that is chemically related to the microRNA of interest selected from the group consisting of an RNA other than a microRNA and a DNA.

35. (Withdrawn) The method of claim 33, where the sample is from a eukaryote.

36. (Withdrawn) The method of claim 33, where the sample is from a primate.

37. (Withdrawn) The method of claim 33, where the sample is from a human.

38. (Withdrawn) The method of claim 33, where the sample comprises a tissue or fluid selected from the group consisting of blood, brain, heart, intestine, liver, lung, pancreas, muscle, a leaf, a flower, a plant root and a plant stem.



39. (Withdrawn) The method of claim 33, where the microRNA of interest consists of 18 or 19 or 20 or 21 or 22 or 23 or 24 RNA residues.

40. (Withdrawn) The method of claim 33, where the microRNA of interest is listed in a public database.

41. (Withdrawn) The method of claim 33, where the sample provided comprises a plurality of microRNAs of interest; and

where each of the plurality of microRNAs of interest has microRNA of interest sequences that are identical to one another.

42. (Withdrawn) The method of claim 33, where the sample provided comprises a plurality of microRNAs of interest comprising a first microRNA of interest having a first microRNA of interest sequence, and a second microRNA of interest having a second microRNA of interest sequence; and

where the first microRNA of interest sequence is different from the second microRNA of interest sequence.

43. (Withdrawn) The method of claim 33, where the sample provided comprises a plurality of microRNAs of interest comprising a first microRNA of interest having a first microRNA of interest sequence, a second microRNA of interest having a second microRNA of interest sequence, and a third microRNA of interest having a third microRNA of interest sequence;

where the first microRNA of interest sequence is different from the second microRNA of interest sequence;

where the first microRNA of interest sequence is different from the third microRNA of interest sequence; and

where second microRNA of interest sequence is different from the third microRNA of interest sequence.

44. (Withdrawn) The method of claim 33, further comprising isolating the total RNA



from the sample after providing the sample.

45. (Withdrawn) The method of claim 33, where the capture probe provided is a set of capture probes;

where each of the capture probes comprises identical first adapter segment sequences;

where each of the capture probes of the set of capture probes comprises identical microRNA binding segment sequences; and

where each of the capture probes of the set of capture probes comprises identical second adapter segment sequences.

46. (Withdrawn) The method of claim 33, where the capture probe provided is a set of capture probes;

where the set comprises at least one capture probe comprising a microRNA binding segment that is substantially complementary to, and capable of hybridizing to, each microRNA listed in a single public database.

47. (Withdrawn) The method of claim 33, where the capture probe provided is a set of capture probes;

where the first capture probe and the second capture probe have identical first adapter segment sequences;

where the first capture probe and the second capture probe have identical microRNA binding segment sequences; and

where the first capture probe has a second adapter segment sequence that is different from the second adapter segment sequence of the second capture probe.

48. (Withdrawn) The method of claim 33, where the capture probe provided is a set of capture probes;

where the set comprises a first capture probe and a second capture probe;



where the first capture probe and the second capture probe have identical first adapter segment sequences;

where the first capture probe and the second capture probe have identical second adapter segment sequences; and

where the first capture probe has a microRNA binding segment sequence that is different from the microRNA binding segment sequence of the second capture probe.

49. (Withdrawn) The method of claim 33, where the capture probe provided is a set of capture probes;

where the set comprises a first capture probe and a second capture probe;

where the first capture probe and the second capture probe have identical microRNA binding segment sequences;

where the first capture probe and the second capture probe have identical second adapter segment sequences; and

where the first capture probe has a first adapter segment sequence that is different from the first adapter segment sequence of the second capture probe.

50. (Withdrawn) The method of claim 33, where the capture probe provided is a set of capture probes;

where the set comprises a first capture probe and a second capture probe;

where the first capture probe and the second capture probe have identical first adapter segment sequences;

where the first capture probe has a microRNA binding segment sequence that is different from the microRNA binding segment sequence of the second capture probe; and

where the first capture probe has a second adapter segment sequence that is different from the second adapter segment sequence of the second capture probe.

51. (Withdrawn) The method of claim 33, where the capture probe provided is a set of



capture probes;

where the set comprises a first capture probe and a second capture probe;

where the first capture probe and the second capture probe have identical microRNA binding segment sequences;

where the first capture probe has a first adapter segment sequence that is different from the first adapter segment sequence of the second capture probe; and

where the first capture probe has a second adapter segment sequence that is different from the second adapter segment sequence of the second capture probe.

52. (Withdrawn) The method of claim 33, where the capture probe provided is a set of capture probes;

where the set comprises a first capture probe and a second capture probe;

where the first capture probe and the second capture probe have identical second adapter segment sequences;

where the first capture probe has a first adapter segment sequence that is different from the first adapter segment sequence of the second capture probe; and

where the first capture probe has a microRNA binding segment sequence that is different from the microRNA binding segment sequence of the second capture probe.

53. (Previously presented) The method of claim 33, where the capture probe provided is a set of capture probes;

where the set comprises a first capture probe and a second capture probe;

where the first capture probe has a first adapter segment sequence that is different from the first adapter segment sequence of the second capture probe;

where the first capture probe has a microRNA binding segment sequence that binds to a microRNA of interest that is different from the microRNA binding segment sequence and the microRNA of interest of the second capture probe; and



where the first capture probe has a second adapter segment sequence that is different from the second adapter segment sequence of the second capture probe.

54. (Withdrawn) The method of claim 33, where the capture probe provided is a set of capture probes;

where the set comprises a first capture probe having a first capture probe sequence, a second capture probe having a second capture probe sequence, and a third capture probe having a third capture probe sequence;

where the first capture probe sequence is different from the second capture probe sequence;

where the first capture probe sequence is different from the third capture probe sequence;  
and

where second capture probe sequence is different from the third capture probe sequence.

55. (Withdrawn) The method of claim 33, where the first linker segment and the second linker segment comprise a substance selected from the group consisting of one or more than one type of polynucleotide, one or more than one type of polynucleotide analog, and a combination of one or more than one type of polynucleotide and polynucleotide analog.

56. (Withdrawn) The method of claim 33, where the first linker, or the second linker, or both the first linker and the second linker are resistant to nuclease degradation.

57. (Withdrawn) The method of claim 56, where the first linker, or the second linker, or both the first linker and the second linker comprise nuclease resistant nucleotides.

58. (Withdrawn) The method of claim 56, where the first linker, or the second linker, or both the first linker and the second linker comprise nucleotides with a phosphothioate backbone that render the first linker, or the second linker, or both the first linker and the second linker resistant to nuclease degradation.

59. (Withdrawn) The method of claim 56, where the first linker, or the second linker, or both the first linker and the second linker comprise nuclease resistant nucleotides and comprise



nucleotides with a phosphothioate backbone that render the first linker, or the second linker, or both the first linker and the second linker resistant to nuclease degradation.

60. (Withdrawn) The method of claim 33, where the first linker and the second linker, each comprises between 6 and 50 residues.

61. (Withdrawn) The method of claim 33, where the first linker comprises at least 10 residues, and at least 10 residues at the 3' end of the first linker are exactly the complement of the corresponding residues at or near the 5' end of the first adapter segment.

62. (Withdrawn) The method of claim 33 where the second linker comprises at least 10 residues, and at least 10 residues at the 5' end of the second linker are exactly the complement of the corresponding residues at or near the 3' end of the second adapter segment.

63. (Withdrawn) The method of claim 33, where the 5' end of the first linker, or the 3' end of the second linker, or both the 5' end of the first linker and the 3' end of the second linker comprise a label.

64. (Withdrawn) The method of claim 33, where the 5' end of first linker comprises one or more than one residue that extends beyond the 3' end of the first adapter segment after the first linker hybridizes to the first adapter segment.

65. (Withdrawn) The method of claim 64, where the one or more than one residue of the 5' end of first linker that extends beyond the 3' end of the first adapter segment functions as a primer binding site.

66. (Withdrawn) The method of claim 33, where the 3' end of second linker comprises one or more than one residue that extends beyond the 5' end of the second adapter segment after the second linker hybridizes to the second adapter segment.

67. (Withdrawn) The method of claim 66, where the one or more than one residue of the 3' end of second linker that extends beyond the 5' end of the second adapter segment functions as a primer binding site.

68. (Withdrawn) The method of claim 33, where the sample, the capture probe, the first



linker and the second linker are combined simultaneously.

69. (Withdrawn) The method of claim 33, further comprising adding one or more than one RNase inhibitor to the combination of the sample, the capture probe, the first linker and the second linker.

70. (Withdrawn) The method of claim 33, where the first adapter segment comprises a solid phase binding group, or the second adapter segment comprises a solid phase binding group, or both the first adapter segment comprises a solid phase binding group and the second adapter segment comprises a solid phase binding group; and

where the method further comprises binding the capture probe to a solid phase before or after combining the sample, the capture probe, the first linker and the second linker.

71. (Withdrawn) The method of claim 70, where the solid phase is a plurality of paramagnetic particles.

72. (Withdrawn) The method of claim 70, where the capture probe is bound to a solid phase through the first adapter segment or through the second adapter segment or through both the first adapter segment and the second adapter segment; and

where the method further comprises purifying the capture probes with hybridized first linker, microRNA of interest and second linker-bound to the solid phase by removing non-hybridized first linkers, second linkers and any other substances that are not bound to the solid phase.

73. (Withdrawn) The method of claim 70, where the solid phase is contained in a vessel comprising a surface and a cap, and where purifying comprises applying a magnetic field to attract the solid phase to the surface of the vessel or the cap of the vessel.

74. (Withdrawn) The method of claim 33, where the first linker hybridizes to the first adapter segment at a position where the last residue on the 3' end of the first linker hybridizes to a residue on the first adapter segment that is between 1 residue and 5 residues from the 3' end of the microRNA binding segment.



75. (Withdrawn) The method of claim 33, where the first linker hybridizes to the first adapter segment at a position where the last residue on the 3' end of the first linker hybridizes to a residue on the first adapter segment that is immediately adjacent to the 3' end of the microRNA binding segment.

76. (Withdrawn) The method of claim 33, where the second linker hybridizes to the second adapter segment at a position where the last residue on the 5' end of the second linker hybridizes to a residue on the second adapter segment that is between 1 residue and 5 residues from the 5' end of the microRNA binding segment.

77. (Withdrawn) The method of claim 33, where the second linker hybridizes to the second adapter segment at a position where the last residue on the 5' end of the second linker hybridizes to a residue on the second adapter segment that is immediately adjacent to the 5' end of the microRNA binding segment.

78. (Withdrawn) The method of claim 33, where the method further comprises purifying the complex.

79. (Withdrawn) The method of claim 33, where the complex is bound to a solid phase through the first adapter segment or through the second adapter segment or through both the first adapter segment and the second adapter segment; and

where the method further comprises purifying the complex by removing non-hybridized first linkers, second linkers and any other substances that are not bound to the solid phase.

80. (Canceled)

81. (Withdrawn-previously presented) The method of claim 33, where the first linker, or the second linker, or both the first linker and the second linker comprise nuclease resistant nucleotides, or comprise nucleotides with a phosphothioate backbone that render the first linker, or the second linker, or both the first linker and the second linker resistant to nuclease degradation; and



where purifying the ligated first linker- microRNA of interest-second linker comprises applying DNAase to a solution containing the ligated first linker- microRNA of interest-second linker to destroy any DNA present in the solution.

82. (Withdrawn-previously presented) The method of claim 33, where purifying the ligated first linker- microRNA of interest-second linker comprises circularizing the ligated first linker- microRNA of interest-second linker.

83. (Withdrawn) A method for identifying a microRNA of interest, the method comprising:

- a) isolating the microRNAs according to claim 33; and
- b) sequencing the microRNA of interest portion of the strand of the ligated first linker- microRNA of interest-second linker.

84. (Withdrawn) The method of claim 83, where sequencing comprises subjecting the strand of the ligated first linker- microRNA of interest-second linker to reverse transcription to produce a double stranded product comprising a first strand of the ligated first linker- microRNA of interest-second linker and a second strand that is the complement of the first strand.

85. (Withdrawn) The method of claim 83, where sequencing comprises amplifying the double stranded product to produce amplification products.

86. (Withdrawn) The method of claim 84, where sequencing comprises cloning the amplification products and culturing the amplification products.